- 51. The gene according to claim 28, wherein the protein which has an aromatic acyl group transfer activity is a protein which transfers an aromatic acyl group to the 3 or 5 position of anthocyanin.
- 52. The gene according to claim 42, wherein the gene encodes a protein which transfers an aromatic acyl group to the 3 or 5 position of anthocyanin.

REMARKS

Entry of the foregoing amendment is respectfully requested. By the foregoing amendment, claim 4 has been canceled without prejudice or disclaimer to the subject matter disclosed therein. Claims 1, 5-8 and 28 have been amended to recite that the protein "transfers an aromatic acyl group to flavonoid". Support for this amendment to claims 1, 5-8 and 28 may be found, at the very least, at page 4, line 34, to page 5, line 1. Claims 5-6 and 42-45 have been amended to recite "consensus sequence" rather than "part of the nucleotide sequence". Support for this amendment to claims 5-6 and 42-45 may be found, at the very least, at page 7, lines 10-11. Claim 7 has been amended to recite that the amino acid sequence is at least 30% homologous to the amino acid sequences of SEQ ID No. 1 to 6. Support for this amendment to claim 7 may be found, at the very least, at page 7, lines 19-23. Claim 8 has been amended to recite that the amino acid sequence is at least 69% homologous to the amino acid sequences of SEQ ID No. 1 to 6. Support for this amendment to claim 8 may be found, at the very least, at page 46, line 18 and at page 7, lines 19-25. Support for new claims 46-52 may be found, at the very least, in the claims as

originally filed and at page 25, lines 27-32, at page 29, lines 6-10, at page 32, lines 26-30, at page 36, line 15, and in Examples 20-23. No new matter has been added by the present invention.

The paper copy of the Sequence Listing for the subject application, is by this amendment, added between the last page of the Specification, currently page 49 and the first page of the Claims to replace the Sequence Listing previously filed on October 12, 1999. Please amend the page numbers accordingly.

In order to expedite prosecution of the present application, applicants wish to respond to the rejections set forth in the Official Action mailed on February 10, 2000 in application Serial No. 08/894,356.

Claims 1-12, 20 and 22-45 have been rejected under 35 U.S.C. § 112, first paragraph, for purportedly not containing a written description of the claimed invention. According to the Examiner, the isolated nucleotide sequences of the present invention encode a single enzyme from a number of plant species, but the claims are purportedly much broader that the scope of the specification. The claims have been amended to recite that the gene encodes a protein which transfers an aromatic acyl group to flavonoid, which of course is a pigment in plants. "Flavonoid" is fully supported throughout the specification.

The following tables summarize the data provided in the specification.

Table A

Example No.	Origin	Clone	Substrate Specificity	
3	Gentians	pGAT4	Transfers to 5-position	
6	Gentians	pGAT106		
8	Petunias	pPAT48		
11	Perillas	pSAT208	Transfer to 3-position	
12	Cinerarias	pCAT8		
20	Lavenders	pLAT21	Transfer to 3-position	

Table B

1.00	pGAT4(5)	pGAT106	pPAT48	pSAT208	pCAT8	pLAT21
pGAT4		38(1)	20(2)	37 ⁽³⁾	28(4)	38 ⁽⁵⁾
pGAT106			16(2)	29(3)	35(4)	37 ⁽⁵⁾
pPAT48				15 ⁽³⁾	16(4)	19 ⁽⁵⁾
pSAT208					37 ⁽⁴⁾	68 ⁽⁵⁾
pCAT8						37 ⁽⁵⁾
pLAT21						

- (1) See page 29, line 1.
- (2) See page 32, line 5.
- (3) See page 36, line 33.
- (4) See page 37, line 35.
- (5) See page 46, line 18.

In Table A, the relationship between clones obtained from various plant origins and the substrate specificity of these clones is described. In Table B, the amino acid homology

between clones is described. As can be seen from the above, the amino acid sequence homology between the proteins encoded by the genes cloned in the present invention is low, despite the fact that these proteins have a common activity. Therefore, the cloned genes encode enzymes which are structurally different, but have a common activity.

Furthermore, the specification does provide guidance as to how one of skill in the art would isolate genes which encode enzymes which have the same activity as the enzymes encoded by the nucleotide sequences set forth in SEQ ID NOS: 1 to 6. For example, the applicants originally isolated a gene which encoded an acyltransferase from the petals of gentians. From this gene, primers were developed and used to develop probes from the discovered gene. These probes were used in a cDNA library to pull out other clones. The plasmid DNA recovered from the cDNA library are separated and their nucleotide sequences are determined. Then, amino acid sequences were obtained from analysis of the purified and isolated nucleotide sequences. The amino acid sequences of the acyltransferases deduced from the DNA nucleotide sequence are compared to confirm that the positive clones are the desired cDNA clone. See page 5, line 31, to page 6, line 24. See also example 3, wherein an acetyltransferase cDNA was isolated from gentians.

At the very least, newly added claims 46 to 52 are allowable. Claims 46-52 specify that the protein is "a protein which transfers an aromatic acyl group to the 3 or 5 position of anthocyanin". These claims find support throughout the specification, and clarify what the activity of the proteins encoded by the claimed genes are.

Claims 1-12, 20 and 22-27 have also been rejected under 35 U.S.C. § 112, first paragraph, because the claims are purportedly not enabled by the specification.

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Specifically, the Examiner purports that the specification only enables claims to the nucleotide sequences depicted in SEQ ID NOS:1 to 6, and the construction and introduction of the acyltransferase gene into petunia. As stated more completely above, the specification does enable claims to nucleotide sequences, including nucleotide sequences other than those depicted in SEQ ID NOS: 1 to 6, which encode a protein which transfers an aromatic acyl group to flavonoid. Specifically, as discussed above, the specification describes how one of skill in the art could obtain other nucleotide sequences from a variety of plant species, wherein said nucleotide sequences encode a protein which transfers an aromatic acyl group to flavonoid.

Furthermore, from the teachings in the specification and known in the art, one of skill in the art would be able to produce a plant whose color has been controlled by introducing thereinto a gene according to claim 1. Techniques for introduction of a hterologous gene into a plant cell, and regeneration of a transgenic plant, has been accomplished prior to the priority date of the present application, and the specification discloses a transgenic petunia, and various genes which could be introduced into plants. Therefore, one of skill in the art could construct a transgenic plant of any species using the genes of the claimed invention.

With regards to the enablement of claims 7 and 8, claim 7 has been amended to restrict the homology to "at least 30%" and claim 8 has been amended to restrict the homology to "at least 68%". Example 3 in the specification describes a gene of gentian origin which encodes a protein, having the amino acid sequence depicted in SEQ ID NO:

1, which transfers an aromatic acyl group to the 5 position of anthocyanin. After filing the

present application, one of the applicants cloned a gene of <u>Torenia hyrida</u> origin encoding a protein, having the amino acid sequence depicted in the attached sequence report, which transfers an aromatic acyl group to the position 5 of anthocyanin. The amino acid sequence homology between the sequence of SEQ ID NO: 1 and that of Trenia 5AT, both of which encode enzymes having the same activity (i.e. they both transfer an aromatic acyl group to the position 5 of flavonoid), is 35%. Therefore, the homology of "at least 30%" in claims should satisfy the enablement requirement. In addition, Example 11 describes a gene of perillas origin, and Example 20 describes a gene of lavender origin. Both the genes encode a protein which transfers an aryl acyl group to the position 3 of flavonoid. The amino acid homology between these two enzymes is 69% (see above Table B and page 46, line 18, of the specification). Therefore, claim 8, which refers to the homology of "at least 69%", is enabled by the specification.

Regarding the enablement of the hybridization type claims (claims 5, 6 and 28), the cDNA cloned in Example 3 (SEQ ID NO: 1) is of gentian origin, and another cDNA (SEQ ID NO: 2) of gentian origin was cloned in Example 6 using a probe developed from the cDNA cloned in Example 3 (see page 28, lines 5 to 18). In addition, in Example 8, a cDNA (SEQ ID NO: 3) of petunia origin was cloned using as a probe a section of the cDNA cloned in Example 3 (see page 31, lines 4 to 8). Further, in Example 11, a cDNA (SEQ ID NO: 8) of perillas origin was cloned using as a primer a conserved sequence of SEQ ID NOS: 1, 3 and 4. Therefore, cDNAs encoding proteins which can transfer an aryl acyl group to flavonoid can be cloned using as a primer or probe a sequence of one of SEQ ID NOS: 1 to 6.

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In light of these remarks, it is believed that the Examiner's rejections under 35 U.S.C. § 112, first paragraph, are moot.

Claims 4, 6, 22, 25 and 33 have been rejected under 35 U.S.C. § 112, second paragraph for purportedly being indefinite. Applicants provide the following remarks regarding these rejections.

Claim 4 has been canceled, rendering its rejection moot.

Claim 6 has been amended to delete "with part" and insert in place thereof "a consensus region" (the same amendment has been made to claim 5). The consensus region is the same as the "conserved" region on page 34, line 30. The meanings of "consensus region" and "conserved region" are established and well known to one of skill in the art. As can be seen from the description on page 34, lines 25-29, the term "conserved region" or "consensus region" means a region in an nucleotide sequence which shares a common sequence with one or more other nucleotide sequences. In the present case, the nucleotide sequences of SEQ ID NOS: 1, 2 and 3 were cloned in Examples 3, 6 and 9, respectively, the amino acid sequence determined, and a conserved (or consensus) amino acid sequence is shown in SEQ ID NO: 21, which was determined by comparison of the amino acid sequences of SEQ ID NOS: 1, 2 and 3. On the basis of the sequence of SEQ ID NO: 21, a primer, SEQ ID NO: 22, was prepared and used to clone another gene encoding a protein which transfers an aromatic acyl group to flavonoid. See Example 11, page 34.

Therefore, claims 5 and 6 satisfy the requirements of 35 U.S.C. § 112, second paragraph.

Claims 22 and 33 have been rejected under 35 U.S.C. § 112, second paragraph, for purportedly being indefinite. The Examiner purports that these claims are redundant.

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Claim 22 depends from claim 1, whereas claim 33 depends from claim 28. The scope of claim 22 is much broader than the scope of claim 33, and thus the claims are not redundant.

Claim 25 has been rejected under 35 U.S.C. § 112, second paragraph, for purportedly being indefinite. Applicants assert that it is clear from claim 25 that what is being claimed is a plant whose color has been controlled or the progeny of said plant, wherein said progeny has its color controlled. Thus, applicants assert that claim 25 is not indefinite.

In light of the above remarks, applicants respectfully submit that all of the previous rejections of the claims are moot. Prompt allowance of the pending claims is believed to be next in order, and such action is earnestly solicited.

In the event that there are any questions relating to this application, the Examiner is

invited to telephone the undersigned so that prosecution of the subject application may be

expedited.

Respectfully submitted,

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